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(54) Title: SINGLE-BATH PREPARATION OF CELLULOSIC MATERIALS

(57) Abstract: The present invention provides methods for single-bath desizing, scouring and bleaching of cellulosic materials, which are carried out by contracting the cellulosic simultaneously or sequentially with an enzyme system and a peracid bleaching system under conditions that do not require emptying the bath or rinsing the cellulosic materials between desizing, scouring and bleaching steps.

SINGLE-BATH PREPARATION OF CELLULOSIC MATERIALS

Field of the Invention

The present invention relates to methods and compositions for treating cellulosic materials, and more specifically, to methods and compositions for desizing, scouring and bleaching cellulosic materials.

Background of the Invention

The processing of cellulosic material, such as cotton fiber, into a material ready for garment manufacture involves several steps: spinning of the fiber into a yarn; construction of woven or knit fabric from the yarn; and subsequent preparation, dyeing and finishing operations. The preparation process, which may involve desizing (for woven goods), scouring, and bleaching, produces a textile suitable for dyeing or finishing.

A. Desizing: Woven goods are the prevalent form of textile fabric construction. The weaving process requires a "sizing" of the warp yarn to protect it from abrasion. Starch, polyvinyl alcohol, carboxymethyl cellulose, waxes and acrylic binders are examples of typical sizing agents commonly used in the industry. In order to ensure a high whiteness and/or a good dyeability, the size and other applied must be thoroughly removed. It is generally believed that an efficient desizing is crucial to the subsequent preparation processes, namely, the scouring and bleaching processes. The sized fabric, in either rope or open width form, is contacted with the processing liquid containing the desizing agent. The desizing agent employed depends upon the type of size to be removed. The most common sizing agent for cotton fabric is based upon starch. Therefore, most often, woven cotton fabrics are desized by a combination of hot water, the enzyme alpha amylase and a wetting agent or surfactant.

B. Scouring: The scouring process removes much of the non-cellulosic compounds naturally found in cotton. In addition to the natural non-cellulosic impurities, scouring can remove residual manufacturing materials introduced, such as spinning, coning or slashing lubricants. Conventional scouring processes typically utilize highly alkaline chemical treatment, which results not only in removal of impurities but also in weakening of the underlying cellulose component of the fiber or fabric. The chemical scouring is followed by extensive rinsing to reduce the risk of re-depositing impurities. Insufficient rinsing yields alkaline residue and uneven removal of impurities

on the fabric, which in turn results in uneven dyeing in the subsequent process. Furthermore, chemical scouring creates environmental problems in effluent disposal, due to the chemicals employed and the materials extracted from the fibers. Enzymes have been proposed as an alternative to conventional chemical agents for scouring cellulosic materials. See, e.g., WO 9824965, WO 0071808, JP 6220772, JP 10088472, U.S. Patent No. 5,912,407; Hartzell et al., *Textile Res.* 68:233 (1998); Hsieh et al., *Textile Res.* 69:590 (1999); Buchert et al., *Text. Chem. Col. & Am. Dyestuff Reptr.* 32:48 (2000); and Li et al., *Text. Chem. Color.* 29:71 (1997).

C. Bleaching: Bleaching of textiles is the final preparation step in the manufacturing of textile fabrics and garments. The purpose of bleaching is to completely remove colored impurities, improve absorbency, and achieve adequate whiteness and dyeability. The most widely used bleaching process in the textile industry is the alkaline hydrogen peroxide process. A conventional textile bleach bath contains: sodium hydroxide, surfactant, optical brightener, stabilizers, and bleaching agents. Bleaching can be carried out in batch wise, semi-continuous, continuous or discontinuous processes. When enzymes are used in either the desizing or scouring process, in order to obtain consistent, high quality results with commercial quantities of textiles, the desizing and/or scouring steps have traditionally been performed separately from the bleaching step because of the high temperature and alkalinity requirement of alkaline peroxide bleaching.

Summary of the Invention

The present invention provides methods for single-bath desizing, scouring and bleaching of cellulosic materials, such as, for example, crude fibers, yarn, or woven or knit textiles, made of cotton, linen, flax, ramie, rayon, hemp, jute, or blends of these fibers with each other or with other natural or synthetic fibers.

The methods of the present invention are carried out by contacting cellulosic materials with (i) an enzyme system and (ii) a bleaching system; by adding the enzyme system and the bleaching system in the same solution containing the cellulosic material to be treated without emptying the bath or rinsing the cellulosic materials between the enzymatic treatment and bleaching steps, *i.e.*, in a single-bath process. The enzyme system and the bleaching system may be added simultaneously to the solution. Alternatively, the enzyme system and the bleaching system may be added sequentially to the solution, in which the cellulosic materials are (i) contacted with the enzyme system for a sufficient time and under appropriate conditions that result in effective bioscouring and/or desizing of the cellulosic material, after which (ii) the bleaching system is added directly to the solution

containing cellulosic materials and the enzyme system, that is without emptying the bath or rinsing the cellulosic materials.

In one aspect of the present invention, methods for treating cellulosic material are disclosed, comprising contacting a cellulosic material with (i) an enzyme system for scouring and/or desizing and (ii) a bleaching system comprising at least one peracid bleaching compound, wherein the enzyme system and the peracid bleaching system are added to the same solution in a single-bath process.

In one embodiment of this aspect of present invention, the enzyme system and the peracid bleaching system are added sequentially in the single-bath process by first (I) contacting a solution containing the cellulosic material with the enzyme system and incubating the solution contents for a sufficient time and under appropriate conditions to promote effective bioscouring and/or desizing, followed by (ii) adding the peracid bleaching system to the same solution containing the cellulosic material and the enzyme system and incubating to complete the processes.

In another embodiment of this aspect of present invention, the enzyme system and the peracid bleaching system are added to the solution containing the cellulosic material simultaneously, *i.e.*, at or about the same time or without an intervening incubation step.

The methods and compositions of present invention provide a product exhibiting a high wettability, high whiteness, and uniformity of mote removal, while having advantages over conventional preparation processes, including: (i) shorter processing times; (ii) conservation of water; and (iii) reduction in waste stream.

Detailed Description of the Invention

According to the present invention, bioscouring and/or desizing are combined with bleaching in a single bath process.

In one preferred embodiment, the single-bath treatment is carried out by adding the enzyme system and the bleaching system simultaneously to the aqueous solution or wash liquor, comprising (i) adding the enzyme system and a peracid bleaching system simultaneously to an aqueous solution or wash liquor which contains or contacts the cellulosic materials, and (ii) incubating for sufficient time and under appropriate conditions to achieve effective scouring and/or desizing, depending on the enzyme system selected, as well as effective bleaching.

In another preferred embodiment, the single-bath treatment is carried out by adding the enzyme system and the bleaching system sequentially to the aqueous solution or wash liquor, comprising (i) adding the enzyme system to an aqueous solution or wash liquor which contains or contacts the cellulosic material; (ii) performing a first incubation for sufficient time and under appropriate conditions to initiate and

cause effective scouring and/or desizing, depending on the enzyme system selected, and (iii) adding the peracid bleaching system to the same solution containing the cellulosic material and the enzyme system and incubating to complete the processes.

Cellulosic Materials:

As used herein, a "cellulosic material" refers to the cellulosic substrate to be treated and includes, for example, cotton, linen, flax, ramie, rayon, hemp, jute, and their blends with other natural or synthetic fibers. The cellulosic materials may also include, for example, crude fiber, yarn, woven or knit textile or fabric, or a garment or finished product.

Enzyme Systems:

As used in the present invention, an "enzyme system" refers to a bioscouring enzyme system and/or a desizing enzyme system. Accordingly, an enzyme system may comprise one or more bioscouring enzymes with or without one or more desizing enzymes or one or more desizing enzymes with or without one or more bioscouring enzymes.

Desizing Enzymes:

Any suitable desizing enzyme may be used in the present invention. Preferably, the desizing enzyme is an amylolytic enzyme. More preferably, the desizing enzyme is an alpha or beta amylase and combinations thereof.

Alpha and beta amylases which are appropriate in the context of the present invention include those of bacterial or fungal origin. Chemically or genetically modified mutants of such amylases are also included in this connection. Preferred alpha-amylases include, for example, alpha-amylases obtainable from Bacillus species, in particular a special strain of *B. licheniformis*, described in more detail in GB 1296839. More preferred amylases include Duramyl™, Termamyl™, Fungamyl™ and BAN™ (all available from Novozymes A/S, Bagsvaerd, Denmark), and Rapidase™ and Maxamyl™ (available from Gist-Brocades, Holland). Other preferred amylolytic enzymes are CGTases (cyclodextrin glucanotransferases, EC 2.4.1.19), e.g., those obtained from species of Bacillus, Thermoanaerobactor or Thermoanaero-bacterium.

The desizing enzymes may also preferably be derived from the enzymes listed above in which one or more amino acids have been added, deleted, or substituted, including hybrid polypeptides, so long as the resulting polypeptides exhibit desizing activity. Such variants useful in practicing the present invention can be created using conventional mutagenesis procedures and identified using, e.g., high-throughput screening techniques such as the agar plate screening procedure.

The desizing enzyme is added to the aqueous solution or wash liquor (i.e., the treating composition) in an amount effective to desize the cellulosic materials. Typically, desizing enzymes, such as alpha-amylases, are incorporated into the treating composition in amount from 0.00001% to 2% of enzyme protein by weight of the composition, preferably in an amount from 0.0001% to 1% of enzyme protein by weight of the composition, more preferably in an amount from 0.001% to 0.5% of enzyme protein by weight of the composition, and even more preferably in an amount from 0.01% to 0.2% of enzyme protein by weight of the composition. The desizing enzyme is preferably used at a level from about 2 to 30,000 KNU/l, more preferably 20-30,000 KNU/l and most preferably 200-300 KNU/l or from about 3-50,000 NAU/l, more preferably 30-5,000 NAU/l, most preferably 350-500 NAU/l.

Bioscouring Enzymes:

Any suitable bioscouring enzyme may be used in the present invention. Preferred bioscouring enzymes include, without limitation, pectinases, proteases, lipases, cutinases and combinations thereof, more preferably, the bioscouring enzyme is a pectinases, and even more preferably, the bioscouring enzyme is a pectate lyase.

Pectinases: Any pectinolytic enzyme composition with the ability to degrade the pectin composition of plant cell walls may be used in practicing the present invention. Suitable pectinases include, without limitation, those of fungal or bacterial origin. Chemically or genetically modified pectinases are also encompassed. Preferably, the pectinases used in the invention are recombinantly produced and are mono-component enzymes.

Pectinases can be classified according to their preferential substrate, highly methyl-esterified pectin or low methyl-esterified pectin and polygalacturonic acid (pectate), and their reaction mechanism, beta-elimination or hydrolysis. Pectinases can be mainly endo-acting, cutting the polymer at random sites within the chain to give a mixture of oligomers, or they may be exo-acting, attacking from one end of the polymer and producing monomers or dimers. Several pectinase activities acting on the smooth regions of pectin are included in the classification of enzymes provided by Enzyme Nomenclature (1992), e.g., pectate lyase (EC 4.2.2.2), pectin lyase (EC 4.2.2.10), polygalacturonase (EC 3.2.1.15), exo-polygalacturonase (EC 3.2.1.67), exo-polygalacturonate lyase (EC 4.2.2.9) and exo-poly-alpha-galacturonosidase (EC 3.2.1.82).

In preferred embodiments of the present invention, the pectinase is a pectate lyase. Pectate lyase enzymatic activity as used herein refers to catalysis of the random cleavage of α-1,4-glycosidic linkages in pectic acid (also called polygalcturonic acid) by

transelimination. Pectate lyases are also termed polygalacturonate lyases and poly(1,4-α-D-galacturonide) lyases.

Any pectate lyase may be used in practicing the present invention. In preferred embodiments, the methods utilize a pectate lyase that exhibits maximal activity at temperatures above about 70°C. Pectate lyases may also preferably exhibit maximal activity at a pH above about 8 and/or exhibit enzymatic activity in the absence of added divalent cations, such as, calcium ions. Non-limiting examples of pectate lyases for use in the present invention include pectate lyases that have been cloned from different bacterial genera such as *Erwinia*, *Pseudomonas*, *Klebsiella* and *Xanthomonas*, as well as from *Bacillus subtilis* (Nasser et al. (1993) *FEBS Letts.* 335:319-326) and *Bacillus* sp. YA-14 (Kim et al. (1994) *Biosci. Biotech. Biochem.* 58:947-949). Purification of pectate lyases with maximum activity in the pH range of 8-10 produced by *Bacillus pumilus* (Dave and Vaughn (1971) *J. Bacteriol.* 108:166-174), *B. polymyxa* (Nagel and Vaughn (1961) *Arch. Biochem. Biophys.* 93:344-352), *B. stearothermophilus* (Karbassi and Vaughn (1980) *Can. J. Microbiol.* 26:377-384), *Bacillus* sp. (Hasegawa and Nagel (1966) *J. Food Sci.* 31:838-845) and *Bacillus* sp. RK9 (Kelly and Fogarty (1978) *Can. J. Microbiol.* 24:1164-1172) have also been described. Any of the above, as well as divalent cation-independent and/or thermostable pectate lyases, may be used in practicing the invention. In preferred embodiments, the pectate lyase comprises the amino acid sequence of a pectate lyase disclosed in Heffron et al., (1995) *Mol. Plant-Microbe Interact.* 8: 331-334 and Henrissat et al., (1995) *Plant Physiol.* 107: 963-976..

The pectinases may be incorporated in the aqueous enzyme solution or wash liquor in an amount from 0.00001% to 2% of enzyme protein by weight of the composition, preferably in an amount from 0.0001% to 1% of enzyme protein by weight of the composition, more preferably in an amount from 0.001% to 0.5% of enzyme protein be weight to the composition, and even more preferably in an amount from 0.01% to 0.2% of enzyme protein by weight of the composition. Pectinases are preferably used at a level from about 2.5 to 500,000 APSU/g fabric, more preferably, at a level from about 25 to 50,000 APSU/g fabric, and most preferably at a level from about 250 to 5,000 APSU/g fabric.

Proteases: Any protease suitable for use in the present invention may be employed. Suitable proteases include those of animal, vegetable or microbial origin, preferably of microbial origin. Preferably, the protease may be a serine protease or a metalloprotease, more preferably, an alkaline microbial protease or a trypsin-like protease. Examples of proteases include aminopeptidases, including prolyl aminopeptidase (3.4.11.5), X-pro aminopeptidase (3.4.11.9), bacterial leucyl aminopeptidase (3.4.11.10), thermophilic aminopeptidase (3.4.11.12), lysyl

aminopeptidase (3.4.11.15), tryptophanyl aminopeptidase (3.4.11.17), and methionyl aminopeptidase (3.4.11.18); serine endopeptidases, including chymotrypsin (3.4.21.1), trypsin (3.4.21.4), cucumisin (3.4.21.25), brachyurin (3.4.21.32), cerevisin (3.4.21.48) and subtilisin (3.4.21.62); cysteine endopeptidases, including papain (3.4.22.2), ficain (3.4.22.3), chymopapain (3.4.22.6), asclepain (3.4.22.7), actinidain (3.4.22.14), caricain (3.4.22.30) and ananain (3.4.22.31); aspartic endopeptidases, including pepsin A (3.4.23.1), Aspergillopepsin I (3.4.23.18), Penicillopepsin (3.4.23.20) and Saccharopepsin (3.4.23.25); and metalloendopeptidases, including Bacillolysin (3.4.24.28).

Commercially available proteases include Alcalase, Savinase, Primase, Duralase, Esperase, Kannase, and Durazym (available from Novozymes A/S), Maxatase, Maxacal, Maxapem, Properase, Purafect, Purafect OxP, FN2, FN3 and FN4 (available from Genencor International Inc.).

Also useful in the present invention are protease variants, such as those disclosed in EP 130,756 (Genentech), EP 214,435 (Henkel), WO 87/04461 (Amgen), WO 87/05050 (Genex), EP 251.446 (Genencor), EP 260.105 (Genencor), Thomas et al., (1985), Nature. 318, p. 375-376, Thomas et al., (1987), J. Mol. Biol., 193, pp. 803-813, Russel et al., (1987), Nature, 328, p. 496-500, WO 88/08028 (Genex), WO 88/08033 (Amgen), WO 89/06279 (Novozymes A/S), WO 91/00345 (Novozymes A/S), EP 525 610 (Solvay) and WO 94/02618 (Gist-Brocades N.V.). The activity of proteases can be determined as described in "Methods of Enzymatic Analysis", third edition, 1984, Verlag Chemie, Weinheim, vol. 5.

Proteases are preferably incorporated into the aqueous enzyme solution or wash liquor in an amount from 0.00001% to 2% of enzyme protein by weight of the composition, preferably in an amount from 0.0001% to 1% of enzyme protein by weight of the composition, more preferably in an amount from 0.001% to 0.5% of enzyme protein be weight to the composition, and even more preferably in an amount from 0.01% to 0.2% of enzyme protein by weight of the composition.

Lipases: Any lipase suitable for use in the present invention may be used. Suitable lipases (also termed carboxylic ester hydrolases) preferably include those of bacterial or fungal origin, including triacylglycerol lipases (3.1.1.3) and Phospholipase A_2.(3.1.1.4.). Lipases for use in the present invention include, without limitation, lipases from *Humicola* (synonym *Thermomyces*), such as from *H. lanuginosa* (*T. lanuginosus*) as described in EP 258 068 and EP 305 216 or from *H. insolens* as described in WO 96/13580; a *Pseudomonas* lipase, such as from *P. alcaligenes* or *P. pseudoalcaligenes* (EP 218 272), *P. cepacia* (EP 331 376), *P. stutzeri* (GB 1,372,034), *P. fluorescens*, *Pseudomonas sp.* strain SD 705 (WO 95/06720 and WO 96/27002), *P. wisconsinensis*

(WO 96/12012); a *Bacillus* lipase, such as from *B. subtilis* (Dartois et al., *Biochem.Biophys. Acta*, 1131:253-360, 1993); *B. stearothermophilus* (JP 64/744992) or *B. pumilus* (WO 91/16422). Other examples are lipase variants such as those described in WO 92/05249, WO 94/01541, EP 407 225, EP 260 105, WO 95/35381, WO 96/00292, WO 95/30744, WO 94/25578, WO 95/14783, WO 95/22615, WO 97/04079 and WO 97/07202. Preferred commercially available lipase enzymes include Lipolase™ and Lipolase Ultra™, Lipozyme™ , Palatase™, Novozym™435, and Lecitase™ (all available from Novovozymes A/S). The activity of the lipase can be determined as described in "Methods of Enzymatic Analysis", Third Edition, 1984, Verlag Chemie, Weinhein, vol. 4.

Lipases are preferably incorporated in the aqueous enzyme solution or wash liquor in an amount from 0.00001% to 2% of enzyme protein by weight of the composition, preferably in an amount from 0.0001% to 1% of enzyme protein by weight of the composition, more preferably in an amount from 0.001% to 0.5% of enzyme protein be weight to the composition, and even more preferably in an amount from 0.01% to 0.2% of enzyme protein by weight of the composition.

Cutinases: Any cutinase suitable for use in the present invention may be used, including, for example, the cutinase derived from *Humicola insolens* cutinase strain DSM 1800, as described in Example 2 of U.S. Patent No. 4,810,414.

Cutinases are preferably incorporated in the aqueous enzyme solution in an amount from 0.00001% to 2% of enzyme protein by weight of the composition, preferably in an amount from 0.0001% to 1% of enzyme protein by weight of the composition, more preferably in an amount from 0.001% to 0.5% of enzyme protein be weight to the composition, and even more preferably in an amount from 0.01% to 0.2% of enzyme protein by weight of the composition.

Suitable bioscouring enzymes also include, for example, bioscouring enzymes derived from the enzymes listed above in which one or more amino acids have been added, deleted, or substituted, including hybrid polypeptides, may be used, so long as the resulting polypeptides exhibit bioscouring activity. Such variants useful in practicing the present invention can be created using conventional mutagenesis procedures and identified using, e.g., high-throughput screening techniques such as the agar plate screening procedure. For example, pectate lyase activity may be measured by applying a test solution to 4 mm holes punched out in agar plates (such as, for example, LB agar), containing 0.7% w/v sodium polygalacturonate (Sigma P 1879). The plates are then incubated for 6 h at a particular temperature (such as, e.g., 75°C). The plates are then soaked in either (i) 1M $CaCl_2$ for 0.5h or (ii) 1% mixed alkyl trimethylammonium Br (MTAB, Sigma M-7635) for 1 h. Both of these procedures cause

the precipitation of polygalacturonate within the agar. Pectate lyase activity can be detected by the appearance of clear zones within a background of precipitated polygalacturonate. Sensitivity of the assay is calibrated using dilutions of a standard preparation of pectate lyase.

Effective scouring typically results in improvement in wettability, when measured using the drop test according to AATCC Test Method 39-1980. Preferably, the wettability of the bleached fabric is 20 seconds or less, most preferably, 10 seconds or less.

Desizing and bioscouring enzymes for use in the invention may be derived from their cell of origin or may be recombinantly produced, and may be purified or isolated. As used herein, a "purified" or "isolated" enzyme is one that has been treated to remove non-enzyme material or other enzymes derived from the cell in which it was synthesized that could interfere with its enzymatic activity. Typically, the desizing and bioscouring enzyme is separated from the bacterial or fungal microorganism in which it is produced as an endogenous constituent or as a recombinant product. If the enzyme is secreted into the culture medium, purification may comprise separating the culture medium from the biomass by centrifugation, filtration, or precipitation, using conventional methods. Alternatively, the enzyme may be released from the host cell by cell disruption and separation of the biomass. In some cases, further purification may be achieved by conventional protein purification methods, including without limitation ammonium sulfate precipitation; acid or chaotrope extraction; ion-exchange, molecular sieve, and hydrophobic chromatography, including FPLC and HPLC; preparative isoelectric focusing; and preparative polyacrylamide gel electrophoresis. Alternatively, purification may be achieved using affinity chromatography, including immunoaffinity chromatography. For example, hybrid recombinant pectate lyases may be used having an additional amino acid sequence that serves as an affinity "tag", which facilitates purification using an appropriate solid-phase matrix.

The desizing and bioscouring enzyme used in the methods of the invention may also be chemically modified to enhance one or more properties that render them even more advantageous, such as, e.g., increasing solubility, decreasing lability or divalent ion dependence, etc. The modifications include, without limitation, phosphorylation, acetylation, sulfation, acylation, or other protein modifications known to those skilled in the art.

Bleaching Systems: As used in accordance with the present invention, a "peracid bleaching system" comprises one or more peracid bleaching compounds, and optionally, an alkali agent, and optionally, at least one bleach stabilizer.

Peracid Bleaching Compound: As used in the present invention, a "peracid bleaching compound" or "peroxy acid bleaching compound" is an acid that contains at least one perhydroxyl group (-OOH). Preferably, the peracid bleaching compound is selected from several classes of organic peroxyacid substances. Preferably, the peracid is a performic acid or carboxylic aliphatic peroxyacids containing a single percarboxylic group and a linear or branched saturated alkyl chain of fewer than 11 carbon atoms. Aliphatic carboxylic peroxyacids containing a linear saturated alkyl chain containing fewer than 6 carbon atoms are also preferred. Examples of such peroxyacids are peracetic acid, perpropanoic acid, per-n-butanoic acid and per-n-pentanoic acid. Peracetic acid is particularly preferred because of its effectiveness and the relative simplicity of methods for its preparation.

In another preferred embodiment of the invention, the organic peroxyacid is selected from diperoxycarboxylic acids containing a linear or branched alkyl chain of fewer than 16 carbon atoms and two percarboxylic groups substituted on carbon atoms situated in alpha-omega positions relative to one another. Examples of such peroxyacids are 1,6-hexanediperoxydioic acid, 1,8-octanediperoxydioic acid, 1,10-decanediperoxydioic acid and 1,12-dodecanediperoxydioic acid. In another preferred embodiment of the invention, the organic peroxyacid is selected from aromatic peroxyacids containing at least one percarboxylic group per benzene nucleus. The aromatic peroxyacids containing only a single percarboxylic group per benzene nucleus will be preferably chosen. An example of such a peroxyacid is peroxybenzoic acid. In yet another preferred embodiment of the invention, an organic peroxyacid is substituted by one or more halogen atoms or by any other organic functional substituents, such as, the carbonyl group (ketone, aldehyde or carboxylic acid), the alcohol group, nitrogen-containing groups such as nitrile, nitro, amine and amide groups, and sulphur-containing groups such as sulpho and mercapto groups. An example of such a peroxyacid is peroxymonosulphuric acid.

The peracid bleaching compound is added to the aqueous solution or wash liquor (i.e., the treating composition) in an amount effective to remove colored impurities, improve absorbency, and achieve adequate whiteness and/or dyeability. Preferably, the peracid bleaching compound is added to the treating composition in an amount from about .01 g/l to about 15 g/l of the composition, more preferably about .1 g/l to 10 g/l of the composition, most preferably about .5 g/l to 5 g/l of the composition.

Alkali Agents:

Alkali agents are well known in the art. Preferred alkali agents used in the present invention include, sodium hydroxide, sodium carbonate, sodium bicarbonate,

sodium perborate, sodium sulfide and sodium sulfite. The alkali agents are preferably added to the treating composition in an amount from about .1 g/l to about 10 g/l of the composition, more preferably, in an amount from about .5 to about 5 g/l.

Bleach Stabilizers:

In another preferred embodiment of the present invention, the bleach composition additionally contains one or more bleach stabilizers. The bleach stabilizers preferably comprise agents which are able to adsorb, bind or complex traces of heavy metals. Examples of agents which can be used according to the invention with a bleach-stabilizing action are polyanionic compounds, such as polyphosphates, polycarboxylates, polyhydroxypolycarboxylates, soluble silicates as completely or partially neutralized alkali metal or alkaline earth metal salts, in particular as neutral Na or Mg salts, which are relatively weak bleach stabilizers. Examples of strong bleach stabilizers which can be used according to the invention are complexing agents, such as ethylenediaminetetraacetate (EDTA), diethylenetriaminepentaacetic acid (DTPA), nitrilotriacetic acid (NTA), methyl-glycinediacetic acid (MGDA), .beta.-alaninediacetic acid (ADA), ethylenediamine-N,N'-disuccinate (EDDS) and phosphonates, such as, ethylenediaminetetramethylenephosphonate, diethylenetriaminepentamethylenephosphonate (DTMPA) or hydroxyethylidene-1,1-diphosphonic acid in the form of the acids or as partially or completely neutralized alkali metal salts.

The bleach stabilizer is preferably added to the treating composition in an amount from about .1 to about 5/g liter of the composition, more preferably from about .5 to about 2g/l, and most preferably about 1 g/l.

The bleach composition according to the invention preferably contains at least one bleach stabilizer, and more preferably, at least one of the above mentioned strong bleach stabilizers. Effective bleaching typically results in one or more of the following properties: (i) a desired whiteness (as determined by Ganz whiteness measurement using, e.g., a Macbeth color eye); (ii) a satisfactory uniformity of mote removal (assessed by visual examination); Preferably, the whiteness of the fabric is 50 Ganz units or higher, and most preferably, 60 Ganz units or higher.

Additional components:

In some embodiments of the invention, the aqueous solution or wash liquor further comprises other components, including, without limitation, other enzymes, as well as surfactants, antifoaming agents, lubricants, builder systems, and the like, that

enhance the scouring and/or bleaching processes and/or provide superior effects related to, e.g., strength, resistance to pilling, water absorbency, and dyeability.

Other enzymes suitable for use in the present invention include, without limitation, pectinases, proteases, and lipases as described above; and cellulases. Cellulases are classified in a series of enzyme families encompassing endo- and exo-activities as well as cellobiose hydrolyzing capability. The cellulase used in practicing the present invention may be derived from microorganisms which are known to be capable of producing cellulolytic enzymes, such as, e.g., species of *Humicola, Thermomyces, Bacillus, Trichoderma, Fusarium, Myceliophthora, Phanerochaete, Irpex, Scytalidium, Schizophyllum, Penicillium, Aspergillus*, or *Geotricum*, particularly *Humicola insolens*, *Fusarium oxysporum*, or *Trichoderma reesei*. Non-limiting examples of suitable cellulases are disclosed in U.S. Patent No. 4,435,307; European patent application No. 0 495 257; PCT Patent Application No. WO91/17244; and European Patent Application No. EP-A2-271 004.

The enzymes may be isolated from their cell of origin or may be recombinantly produced, and may be chemically or genetically modified. Typically, the enzymes are incorporated in the aqueous solution at a level of from about 0.0001% to about 1% of enzyme protein by weight of the composition, more preferably from about 0.001% to about 0.5% and most preferably from 0.01% to 0.2%. It will be understood that the amount of enzymatic activity units for each additional enzyme to be used in the methods of the present invention in conjunction with a particular bioscouring enzyme can be easily determined using conventional assays.

Surfactants suitable for use in practicing the present invention include, without limitation, nonionic (see, e.g., U.S. Patent No. 4,565,647); anionic; cationic; and zwitterionic surfactants (see, e.g., U.S. Patent No. 3,929,678); which are typically present at a concentration of between about 0.2% to about 15% by weight, preferably from about 1% to about 10% by weight. Anionic surfactants include, without limitation, linear alkylbenzenesulfonate, α-olefinsulfonate, alkyl sulfate (fatty alcohol sulfate), alcohol ethoxysulfate, secondary alkanesulfonate, alpha-sulfo fatty acid methyl ester, alkyl- or alkenylsuccinic acid, and soap. Non-ionic surfactants include, without limitation, alcohol ethoxylate, nonylphenol ethoxylate, alkylpolyglycoside, alkyldimethylamineoxide, ethoxylated fatty acid monoethanolamide, fatty acid monoethanolamide, polyhydroxy alkyl fatty acid amide, and N-acyl N-alkyl derivatives of glucosamine ("glucamides").

Builder systems include, without limitation, aluminosilicates, silicates, polycarboxylates and fatty acids, materials such as ethylenediamine tetraacetate, and metal ion sequestrants such as aminopolyphosphonates, particularly ethylenediamine

tetramethylene phosphonic acid and diethylene triamine pentamethylenephosphonic acid, which are included at a concentration of between about 5% to 80% by weight, preferably between about 5% and about 30% by weight.

Antifoam agents include without limitation silicones (U.S. Patent No. 3,933,672; DC-544 (Dow Corning), which are typically included at a concentration of between about 0.01% and about 1% by weight.

The compositions may also contain soil-suspending agents, soil-releasing agents, optical brighteners, abrasives, and/or bactericides, as are conventionally known in the art.

Process conditions:

The manner in which the aqueous solution containing the enzyme and bleaching system is contacted with the cellulosic material will depend upon whether the processing regime is continuous, discontinuous pad-batch or batch. For example, for continuous or discontinuous pad-batch processing, the aqueous enzyme solution is preferably contained in a saturator bath and is applied continuously to the cellulosic material as it travels through the bath, during which process the cellulosic material typically absorbs the processing liquor at an amount of 0.5-1.5 times its weight. In batch operations, the fabric is exposed to the enzyme solution for a period ranging from about 5 minutes to 24 hours at a liquor-to-fabric ratio of 5:1-50:1.

The aqueous solution or wash liquor typically has a pH of between about 4 and about 11. Preferably, the pH of the treating composition is between about 5 and about 10, preferably between about 7 to about 9, and most preferably about 8 to about 9. The temperature at which the combined scouring and/or desizing and bleaching processes are carried out will depend on the process used. In the case of cold pad batch process, the scouring and/or desizing and bleaching temperature is preferably between about 15°C and about 45°C, and most preferably between about 25°C and about 35°C. For continuous and other batch processes, the scouring and/or desizing temperature is preferably between about 35°C and about 75°C, and most preferably between about 45°C and about 65°C; and the bleaching temperature may be between about 30°C and about 100°C, preferably between about 50°C and about 100°C, and most preferably between about 60°C and about 90°C.

It will be understood that the optimum dosage and concentration of the enzymes, bleaching compounds, bleach stabilizers, and alkali agents, the volume of the aqueous solution or wash liquor, and the pH and temperature will vary, depending on: (i) the nature of the fiber, i.e., crude fiber, yarn, or textile; (ii) whether simultaneous or sequential scouring and bleaching are carried out; (iii) the particular enzyme(s)

used, and the specific activity of the enzyme; (iv) the conditions of temperature, pH, time, etc., at which the processing occurs; (v) the presence of other components in the wash liquor; and (vi) the type of processing regime used, i.e., continuous, discontinuous pad-batch, or batch. The optimization of the process conditions can be determined using routine experimentation, such as, by establishing a matrix of conditions and testing different points in the matrix. For example, the amount of enzyme, the temperature at which the contacting occurs, and the total time of processing can be varied, after which the resulting cellulosic materials or textile is evaluated for (a) pectin removal; (b) a scoured property such as, e.g., wettability; and (c) quality of bleaching, such as whiteness.

In a preferred embodiment, the conditions or treating composition may be adjusted to favor the desizing, scouring or bleaching processes, such as, by adjusting pH, concentration of wetting agent, or concentration of divalent cationic chelator such as ethylene diamine tetraacetate so as to further promote the bleaching process. In a preferred embodiment, the sequential mode may further comprise adjusting one or more properties of the composition of the aqueous solution or wash liquor between steps (ii) and (iii). For example, pH, concentration of wetting agent, or concentration of divalent cationic chelator, such as, ethylene diamine tetraacetate, may be adjusted between steps (ii) and (iii) so as to further promote the bleaching process. The conditions of the first and second incubations may also differ with respect to temperature, agitation, time, and the like.

The following examples are intended as non-limiting illustrations of the present invention.

Example 1: <u>Single-bath Simultaneous Bioscouring and Bleaching with H_2O_2</u>

A. ***Bioscouring and Bleaching***: A 45 cm x 21.5 cm fabric weighing about 25 gram was cut from an interlock knit fabric (type 4600, Ramseur Co., NC). The fabric was loaded into a Labomat beaker (Mathis Labomat, Werner Mathis USA, Inc, NC), which was then filled with 250 mL of 20mM sodium bicarbonate-carbonate buffer solution (pH9.2) containing 3000 APSU/kg fiber of pectate lyase, 0.5g/l wetting agent (Basophen M, BASF), 1.7g/L H_2O_2, and 0.75g/l stabilizer (sodium silicate). The fabric was treated at 55°C for 15 minutes after which temperature was raised at 5°C /minute to 70°C for 1 hour. The fabric was then washed thoroughly with tap water to remove the residual chemicals and dried at room temperature overnight.

B. ***Analysis***: Whiteness of the fabric was measured by a Macbeth color eye in Ganz units. Wettability was determined by a drop test, measuring the time in seconds for a drop of water to be absorbed by the fabric.

The results are presented in Table 1. Both the whiteness and wettability of the fabric were very low. This example illustrates that bleaching the knitted fabric with hydrogen peroxide alone in the absence of alkali resulted in insufficient improvement in whiteness, wettability and mote removal.

Example 2: Single-bath Simultaneous Bioscouring and Bleaching with H_2O_2/NaOH

The experiment was conducted in essentially the same manner as example 1 above, except that the 2g/l of NaOH was added to the bioscouring/bleaching solution.

The results are shown in Table 1. It is evident that the presence of alkali dramatically improved the whiteness of the fabric. However, the wettability of the fabric is very poor, since alkali may have denatured the enzyme.

Example 3: Single-bath Sequential Bioscouring and Bleaching with H_2O_2/NaOH

A. ***Bioscouring:*** A 45 cm x 21.5 cm fabric weighing about 25 gram was cut from an interlock knit fabric (type 4600, Ramseur Co., NC). The fabric was loaded into a Labomat beaker (Mathis Labomat, Werner Mathis USA, Inc, NC), which was then filled with 250 mL of 20mM sodium bicarbonate-carbonate buffer solution (pH9.2) containing 3000 APSU/kg fiber of pectate lyase and 0.5g/l wetting agent (Basophen M, BASF). The fabric was treated at 55°C for 15 minutes.

B. ***Bleaching***: To the same beaker, add H_2O_2, NaOH and sodium silicate. The final concentrations of H_2O_2, NaOH and sodium silicate were the same as Example 1 above. The Labomat temperature was raised at 5°C /minute to 70°C for 1 hour, after which the water was drained. The fabric was then washed thoroughly with tap water to remove the residual alkali and dried at room temperature.

The results are shown in Table 1. Compared to the simultaneous mode, the wettability of the fabric was improved.

Example 4: Two-bath Bioscouring and Bleaching with H_2O_2/NaOH

The experiment was conducted in essentially the same manner as example 3 above, except that the scouring solution was drained and replaced with water after the bioscouring stage.

The results are shown in Table 1 below. This example demonstrates that the two-bath process mode (example 4) for peroxide bleaching is superior to the one-bath mode (example 3), as evidenced by much higher whiteness, better wettability, and more effective mote removal.

Example 5: Two-bath Scouring and Bleaching with H_2O_2/NaOH

The experiment was conducted in essentially the same manner as example 4 above, except that pectate lyase was absent from the scouring solution.

The results are shown in Table 1. Because of the absence of pectate lyase from the bioscouring solution, the wettability of the fabric was very poor. This example demonstrates that bioscouring enzyme is very important for improving the wettability of the fabric.

Table1. Bioscouring and peroxide bleaching of knitted fabrics

Example #	Process Mode	Scouring	Bleaching	Whiteness,Ganz 82	Wettability, Seconds	Motes
1	Single-bath Simultaneous	Enzyme	Peroxide	44.3	>60	5
2	Single-bath Simultaneous	Enzyme	Peroxide/NaOH	59.2	>60	1
3	Single-bath Sequential	Enzyme	Peroxide/NaOH	58.5	39	2
4	Two-bath	Enzyme	Peroxide/NaOH	63.0	10	1
5	Two-bath	Buffer	Peroxide/NaOH	62.7	>60	1

*Rating of motes: 1: the fewest; 5: the most.

Example 6: Single-bath Simultaneous Bioscouring and Bleaching with Peracetic Acid (PA)

A 45 cm x 21.5 cm fabric weighing about 25 gram was cut from an interlock knit fabric (type 4600, Ramseur Co., NC). The fabric was loaded into a Labomat beaker (Mathis Labomat, Werner Mathis USA, Inc, NC), which was then filled with 250 mL of 20mM sodium phosphate buffer solution (pH9.2) containing 3000 APSU/kg fiber of pectate lyase, 0.5g/l wetting agent (Kierlon Jet B, BASF), 50mM peracetic acid, and 0.75g/l stabilizer (Calgon, Dexter). The fabric was treated at 55°C for 15 minutes after which temperature was raised at 5°C /minute to 70°C for 1 hour. The fabric was then

washed thoroughly with tap water to remove the residual chemicals and dried at room temperature overnight.

The results are shown in Table 2. It is evident that peracetic acid is more effective bleaching agent than peroxide (example 6 vs. 1) under the conditions used in this experiment.

Example 7: Single-bath Sequential Bioscouring and Bleaching with Peracetic Acid (PA)

A. ***Bioscouring:*** A 45 cm x 21.5 cm fabric weighing about 25 gram was cut from an interlock knit fabric (type 4600, Ramseur Co., NC). The fabric was loaded into a Labomat beaker (Mathis Labomat, Werner Mathis USA, Inc, NC), which was then filled with 250 mL of 20mM sodium phosphate buffer solution (pH9.2) containing 3000 APSU/kg fiber of pectate lyase and 0.5g/l wetting agent (Kierlon Jet B, BASF). The fabric was treated at 55□C for 15 minutes.

B. ***Bleaching***: To the same beaker, add peracetic acid and Calgon. The final concentrations of peracetic acid and Calgon were the same as Example 6 above. The Labomat temperature was raised at 5□C /minute to 70□C for 1 hour, after which the water was drained. The fabric was then washed thoroughly with tap water to remove the residual alkali and dried at room temperature.

The results are shown in Table 2. This example demonstrates that one-bath sequential mode for bioscouring and peracetic acid bleaching resulted in much better wettability of the fabric than the simultaneous mode.

Example 8: Single-bath Sequential Scouring and Bleaching with Peracetic Acid (PA)

The experiment was conducted in essentially the same manner as example 7 above, except that pectate lyase was absent from the scouring solution.
The results are shown in Table 2 below. This example further demonstrates that bioscouring enzyme improves the wettability of the fabric.

Example 9: Single-bath Sequential Bioscouring and Bleaching with Peracetic Acid (PA) and NaOH

The experiment was conducted in essentially the same manner as example 7 above, except that 2g/L of NaOH was added to the bleach solution.

The results are shown in Table 2 below. The example illustrates that addition of alkali to the bleach bath further improved whiteness and wettability of the fabric.

Example 10: Two-bath Bioscouring and Bleaching with Peracetic Acid (PA)

The experiment was conducted in essentially the same manner as example 7 above, except that the scouring solution was drained and replaced with water after the bioscouring stage.

The results are shown in Table 2 below. Surprisingly, the whiteness and mote removal of the two-bath process mode (example 9) was not as good as the one-bath sequential mode (example 7), which is very different from what has been observed in bioscouring and peroxide bleaching.

Example 11: Two-bath Scouring and Bleaching with Peracetic Acid (PA)

The experiment was conducted in essentially the same manner as example 10 above, except that pectate lyase was absent from the scouring solution.

The results are shown in Table 2. The wettability of the fabric was very poor. This example further demonstrates that bioscouring enzyme improves the wettability of the fabric.

Table 2. Bioscouring and peracetic acid bleaching of knitted fabrics

Example #	Process Mode	Scouring	Bleaching	Whiteness,Ganz 82	Wettability, Seconds	Motes*
Starting Fabric				8.5	>60	5
6	One-bath Simultaneous	Enzyme	PA	62.6	40	1
7	One-bath Sequential	Enzyme	PA	62.1	4	1
8	One-bath Sequential	Buffer	PA	61.8	26	1
9	One-bath Sequential	Enzyme	PA/NaOH	64.2	2	1
10	Two-bath	Enzyme	PA	59.3	3	2

11	Two-bath	Buffer	PA	59.6	>60	2

* Rating of motes: 1: the fewest; 5: the most.

* PA: Peracetic acid.

Example 12: Single-bath Sequential Bioscouring and Bleaching with H_2O_2/NaOH

The experiment was conducted in essentially the same manner as example 3 above, except that 25 gram of desized woven fabric (428U, Testfabric, INC., PA) was used to replace the knitted fabric.

The results are shown in Table 3. It is clear that sequential bioscouring and peroxide bleaching in a single bath resulted in a woven fabric with very low whiteness.

Example 13: Two-bath Bioscouring and Bleaching with H_2O_2/NaOH

The experiment was conducted in essentially the same manner as example 4 above, except that 25 gram of desized woven fabric (428U, Testfabric, INC., PA) was used to replace the knitted fabric.

The results are shown in Table 3. This example further demonstrates that the two-bath process mode for peroxide bleaching is much better than the one-bath mode.

Example 14: Single-bath Sequential Bioscouring and Bleaching with Peracetic acid (PA)/NaOH

The experiment was conducted in essentially the same manner as example 12 above, except that the bleaching solution contained 50 mM peracetic acid (Aldrich), 2g/L NaOH and 0.75g/L Calgon (BASF).

The results are shown in Table 3. This example demonstrates that peracetic acid (example 14) is more effective bleaching agent than peroxide (example 12).

Example 15: Single-bath Sequential Bioscouring and Bleaching with Peracetic acid (PA)/NaOH

The experiment was conducted in essentially the same manner as example 13 above, except that the bleaching solution contained 50 mM peracetic acid (Aldrich), 2g/L NaOH and 0.75g/L Calgon (BASF).

The results are shown in Table 3. Similar to what have been observed from the knitted fabric examples (6-10), this example demonstrates that the one-bath sequential mode is much better than the two-bath process mode (example 9) in terms of improvement in whiteness and mote removal.

Table 3. Bioscouring and bleaching of woven fabrics

Example #	Process Mode	Scouring	Bleaching	Whiteness,Ganz 82	Wettability, Seconds	Motes
12	Single-bath Sequential	Enzyme	Peroxide/NaOH	50.3	1	2
13	Two-bath	Enzyme	Peroxide/NaOH	55.4	1	1
14	Single-bath Sequential	Enzyme	PA/NaOH	63.3	1	1
15	Two-bath	Enzyme	PA/NaOH	59.5	1	1

*Rating of motes: 1: the fewest; 5: the most.

All patents, patent applications, and literature references referred to herein are hereby incorporated by reference in their entirety. Many variations of the present invention will suggest themselves to those skilled in the art in light of the above detailed description. Such obvious variations are within the full-intended scope of the appended claims.

Claims:

1. A method for treating cellulosic material, comprising contacting the cellulosic material with (i) an enzyme system for desizing and/or bioscouring the cellulosic material and (ii) a bleaching system comprising at least one peracid bleaching compound, wherein the enzyme system and the bleaching system are added simultaneously or sequentially to a single solution containing the cellulosic material.

2. The method of claim 1, wherein the enzyme system and the bleaching system are added simultaneously to the solution containing the cellulosic material.

3. The method of claim 1, wherein the enzyme system and the bleaching system are added sequentially to the solution containing the cellulosic material, comprising (i) adding the enzyme system and incubating, and subsequently (ii) adding the bleaching system and incubating.

4. The method of claim 1, wherein the cellulosic material is contacted with the enzyme system and the bleaching system to produce a fabric with a wettability of 20 seconds or less and whiteness of at least 50 Ganz units.

5. The method of claim 1, wherein the cellulosic material is (i) contacted with the enzyme system to produce a fabric with wettability of 20 seconds or less, after which (ii) the bleaching system is added to the solution containing the cellulosic material.

6. The method of claims 3, further comprising between said (i) and said (ii), adjusting a property of the solution selected from the group consisting of pH, ionic strength, temperature, concentration of surfactant, concentration of divalent cationic chelator, and combinations of any of the foregoing.

7. The method of claim 1, wherein the enzyme system comprises at least one enzyme for desizing the cellulosic material and at least one enzyme for bioscouring the cellulosic material

8. The method of claim 1, wherein the enzyme system is a bioscouring enzyme system.

9. The method of claim 1, wherein the enzyme system is a desizing enzyme system.

10. The method of claim 1, wherein the desizing enzyme system comprises at least one desizing enzyme selected from the group consisting of an alpha-amylase and a beta-amylase, and combinations thereof.

11. The method of claim 1, wherein the bioscouring enzyme system comprises at least one bioscouring enzyme selected from the group consisting of pectinase, protease, lipase, and combinations of any of the foregoing.

12. The method of claim 1, wherein the bioscouring enzyme system comprises at least one bioscouring enzyme selected from the group consisting of pectate lyase, pectin lyase, polygalacturonase, exo-polygalacturonase, exo-polygalacturonate lyase and exo-poly-alpha-galacturonosidase.

13. The method of claim 1, wherein the bioscouring enzyme system comprises a pectate lyase.

14. The method of claim 1, wherein the bioscouring enzyme system comprises a protease selected from the group consisting of aminopeptidases, serine endopeptidases, cysteine endopeptidases, aspartyl endopeptidases, and metalloendopeptidases.

15. The method of claim 1, wherein the bioscouring enzyme system comprises a lipase selected from the group consisting of triacylglycerol lipases and phospholipases.

16. The method of claim 1, wherein the bioscouring enzyme system comprises a pectate lyase that exhibits maximal pectate lyase enzymatic activity at a temperature above about 70°C.

17. The method of claim 1, wherein the bioscouring enzyme system comprises a pectate lyase that exhibits maximal pectate lyase enzymatic activity at a pH above about 8.

18. The method of claim 1, wherein the cellulosic material comprises a textile.

19 The method of claim 1, wherein the cellulosic material comprises cotton.

20. The method of claim 1, wherein said single solution further comprises one or more buffers, surfactants, chelating agents, and/or lubricants, or salts of any of the foregoing.

21. The method of claim 1, wherein the at least one peracid bleaching compound is an organic peroxyacid compound.

22. The method of claim 1, wherein the at least one peracid bleaching compound is peracetic acid.

23. The method of claim 1, wherein the at least one peracid bleaching compound is selected from the following classes of organic peroxyacid substances: performic acid and carboxylic aliphatic peroxyacids; diperoxycarboxylic acids; aromatic peroxyacids; organic peroxyacid substituted by one or more halogen atoms or by any other organic functional group.

24. The method of claim 1, wherein the contacting of the cellulosic material with the at least one peracid bleaching compound is performed with at least one bleach stabilizer.

25. The method of claim 23, wherein the bleach stabilizer is selected from the group consisting of ethylenediaminetetraacetate (EDTA), diethylenetriaminepentaacetic acid (DTPA), nitrilotriacetic acid (NTA), methyl-glycinediacetic acid (MGDA), .beta.-alaninediacetic acid (ADA), ethylenediamine-N,N'-disuccinate (EDDS), ethylenediaminetetramethylenephosphonate, diethylenetriaminepentamethylenephosphonate (DTMPA) or hydroxyethylidene-1,1-diphosphonic acid.

26. The method of claim 1, wherein the contacting of the cellulosic material with the at least one peracid bleaching compound is performed with at least one alkaline agent.

27. The method of claim 26, wherein the alkaline agent is sodium hydroxide.

28. The method as defined in claim 1, wherein the contacting of the cellulosic material with the at least one peracid bleaching compound is performed with at least one bleach stabilizer and at least one alkaline agent.

29. A method for treating a cellulosic material, said method comprising contacting the cellulosic material with (a) at least one bioscouring enzyme and (b) at least one peracid bleaching compound in a single-bath solution comprising the cellulosic material.

30. The method of claim 29, wherein the at least one bioscouring enzyme and the at least one peracid bleaching compound are added simultaneously to the single-bath solution comprising the cellulosic material.

31. The method of claim 29, wherein the at least one bioscouring enzyme and the at least one peracid bleaching compound are added sequentially to the single-bath solution comprising the cellulosic material, said sequential process comprising (i) adding the at least one bioscouring enzyme to the solution comprising the cellulosic material and incubating the cellulosic material and the bioscouring enzyme, followed by (ii) adding the at least one peracid bleaching compound to the solution comprising the cellulosic material and the bioscouring enzyme and incubating.

32. The method of claim 29, further comprising contacting the cellulosic material with at least one desizing enzyme.

33. The method of claim 32, wherein the desizing enzyme is added to the single-bath solution.

34. The method of claim 32, wherein the desizing enzyme is added to the single-bath solution prior to the addition of the bioscouring enzyme.

35. The method of claim 32, wherein the desizing enzyme is added to the single-bath solution simultaneously with the bioscouring enzyme.

36. The method of claim 32, wherein the desizing enzyme is added to the single-bath solution simultaneously with the bioscouring enzyme and the peracid bleaching compound.

37. The method of claim 30, wherein the contacting of the cellulosic material with the at least one desizing enzyme is performed in a separate bath then the single-bath solution used for contacting cellulosic material with the at least one bioscouring enzyme and the at least one peracid bleaching compound.

38. A method for treating a cellulosic material, said method comprising contacting the cellulosic material with (a) at least one bioscouring enzyme, (b) at least one desizing enzyme and (c) at least one peracid bleaching compound in a single-bath solution.

39. The method of claim 38, wherein the at least one bioscouring enzyme, the at least on desizing enzyme, and the at least one peracid bleaching compound are added simultaneously to the single-bath solution comprising the cellulosic material.

40. The method of claim 38, wherein the method comprises adding at least one bioscouring enzyme and the at least one desizing enzyme to the single-bath solution comprising the cellulosic material, incubating the at least one bioscouring enzyme, the at least one desizing enzyme and the cellulosic material, followed by adding at least one peracid bleaching compound and incubating.

41. A method for treating a cellulosic material, said method comprising contacting the cellulosic material with (a) at least one desizing enzyme and (b) at least one peracid bleaching compound in a single-bath solution comprising the cellulosic material.

42. The method of claim 41, wherein the at least one desizing enzyme and the at least one peracid bleaching compound are added simultaneously to the single-bath solution comprising the cellulosic material.

43. The method of claim 41, wherein the at least one desizing enzyme and the at least one peracid bleaching compound are added sequentially to the single-bath solution comprising the cellulosic material, said sequential process comprising (i) adding the at least one desizing enzyme to the solution comprising the cellulosic material and incubating the cellulosic material and the desizing enzyme, followed by (ii) adding the at least one peracid bleaching compound to the solution comprising the cellulosic material and the desizing enzyme and incubating.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US02/20925

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : D06M 23/00
US CL : 8/116.1, 137, 138

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 8/116.1, 137, 138

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
WEST

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 6,162260 A (LIU et al.) 19 December 2000 (19.12.2000), abstract, column 2, lines 5-12, 20-25, 58-67; column 3, lines30-35; column 4, lines 9-15; column 5, lines 64-67; column 6, lines 5-10; column 7, lines 34-67.	1-3 and 5-42
X	US 6,124,127 A (ANDERSEN et al.) 26 September 2000 (26.09.2000), column 3, lines 19-30; column 15, lines22-67; column 16, lines 18-25.	1-42

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

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Date of the actual completion of the international search	Date of mailing of the international search report
09 September 2002 (09.09.2002)	17 SEP 2002
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703)305-3230	Authorized officer Yogendra N. Gupta Telephone No. 703-872-9309

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